Oslo, 23 July 1955.

Professor, Mr. Joshua Lederberg,
Department of Genetics,
University of Wisconsin,
Madison 6, Wisconsin.

Dear prof. Lederberg,

I have read your reprints with great interest. I hope that you have received my reprints, and in them you may have seen that I do not consider the recombination by genophores as an alternative to sexual recombination, but on the contrary I consider it as the origin of the sexual recombination. We may expect to find all intermediate stages between exchange of uncomplete genophores (carrying only a part of the bacteriums chromogenes) and complete genophores or spermnuclei (carrying all the chromogenes) as in Paramecium; as well as we may find intermediate stages between remote transmission of heredity (salmonella) and neighbor-exchange of heredity (K_{12} and Paramecium) and between this and cellular fusion (green algae).

Nevertheless,- I hope you will excuse me ! - the results presented in the reprints you have sent to me and the papers you have published before give, after my opinion, the prove that K_{12} has more than one and probably no less than three genophores. Morover there exist at present no prove that any of this genophores is a complete one, although this may very well be the case.

For the first (phage-genophore λ):

The tempered phage λ is able to transduce not only the gene Lp, but also the gene Gal, if I have well understood your papers. Both of this are chromogenes, that means the phage is a genophore, and an uncomplete one, as most of the chromogenes are not transduced by λ.

For the second (genophore Δ):

Some K_{12} bacteria are shown to be partly diploids and partly haploids. I have not seen any complete record over the "diploidic" genes. But what I have seen is sufficient to state that the partial diploidecity can neither be explained by the adsorption of a λ genophore nor by the emission of a λ genophore from an entirely diploid cell. The only plausible explanation of the phenomenon I have been able to find is that there may exist a second uncomplete genophore Δ. Do you have a better explanation, please let me know.

For the third (X-genophores):

There are still some chromogenes which are exchanged among K_{12} bacteria and which are not carried by Δ genophores nor by λ phage-genophores. We have no reason to postulate the existence of a different crossbreading system for this genes. The most plausible assumption, I think, is that there exist one more or several more genophores. (X-genophore or genophores). X or one of the X's may very well be a complete genophore or a gametic nucleus (if you prefer), but this is a question which must still be decided.

I suppose you may have already in your material enough informations in order to find the range of heredity carried by the λ and Δ genophores. Perhaps it may be more difficult to find the range of heredity carried by the X or the X's.

The existence of several different genophores may give different linking relations among genes than what would be expected in a species with cell fusion or with a single complete genophore or gametic nucleus. Linking relations could appear among genes belonging to different chromosomes if they can be carried by the same genophore, and may illude the existence of a single chromosome containing all the Lederbergian heredity (chromogenes) of the bacterium.

If I should test the genophore-hypothesis on K_{12}, then I would need all available data on linking relations among chromogenes and I would need to know which genes are found to be diploides and which are found to be haploides in the cases of partial diploidy. But may be you prefer to test by yourself the genophore assumption on your data. I would like this solution and in the case you agree I hope you will hold me informed concerning the results you obtain.

My best regards

Nils Aall Barricelli

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N o r w a y

On the experimental test of the genophore-hypotheses for bacterial crosses.

(This paper is conceived as a suggestion of experimental works in order to test the genophore hypotheses.)

In some crosses performed by Lederberg[1] with lysogenic, sensitive and resistent K_{12} strains, the following results were obtained:

Parents			Segregation
1) Immune-2	x	Lysogenic	Parental and sensitive
2) Immune-1	x	Immune-2	" " "
3) Lysogenic-immune-2	x	Sensitive	" , lysogenic and immune-2

The two first crosses does not segregate all the expected varieties. By cross 1 we would expect to obtain not only sensitive, but also lysogenic-immune-2 bacteria. In cross 2 we would expect to obtain besides sensitive also immune 1 and 2 bacteria. The viability of lysogenic-immune-2 is doubtless as the variety is used in cross 3 with sensitive and segregate normality.

This lack of segregation fits very well with the "genophore" hypotheses[2] assuming several uncomplete genophores while it is not so easy to see how it can be fitted with the assumption of a single "gametic nucleus" or "complete genophore" carring all the Lederbergian heredity from an F_+ to an F_- cell[3].

According to the genophore hypotheses we would expect that the hereditary material transmitted from an F_+ to an F_- bacterium need not to be carried by a single complete genophore, but may very well be carried by several uncomplete genophores. For instance we may tentatively assume:

1) A λ-genophore (or phage if virulent)
   carring the genes Lp_1, Gal_1, Gal_2, Gal_3, Gal_4 etc.
2) A Δ-genophore
   carring the genes S and Mal

(1) E. and J. Lederberg. Genetics, Vol. 38, No.1, January 1953.p.51-60.
(2) N.Aall Barricelli - Acta Biotheoretica. Vol. XI, p. II, 1955.
(3) J. Lederberg - Journal of Cellular and Comparative Physiology Vol. 45, Supplement 2, June 1955.

3) <u>One X-genophore (or several x-genofores)</u>
carring the rest of the genes.

The various genophores may rest in a bacterium for shorter or longer time and may possibly reproduce together with the genes of the bacterium for several generations producing some kinds of partial diploidicity. Moreower, the F_- parent need not always receive all the genophores in a mating with an F_+. This may also produce some cases of partial diploidicity and may explaine the relative bias in favor of markers from the F_- parent. The reason for this may for instance be some kind of "immunity" in the F_- parent against one or several genophores or impotence in the F_+ parent to produce one of the genophores.

The case of immunity which have interest for the Lederbergian crossing experiments reported above, is the immunity-2 against the λ-phage. No variety of λ which is known today seems able to invade or induce lysogenity in immune-2 bacteria.

If this is true also for conjugation, that means if an F_- immune-2 conjugant can not receive the λ-genophore from the F_+ parent, then we would have the explanation of the crossing results reported above. We need only to assume that in the experiments 1 and 2 Lederberg has used an F_- immune-2 parent. As immune-2, according to our assumption, can not receive the λ-genophore, it can not segregate the <u>lysogenic-immune-2</u> variety in the first crossing nor the <u>immune 1 and 2</u> variety in the second crossing.

In the third cross on the contrary the <u>lysogenic-immune-2</u> parent used by Lederberg was obtained by λ_2-selection from the F_+- lysogenic parent used in the first cross. This was therefore an F_+. The F_- parent was the sensitive one and could very well receive a λ-genophore and segregate a lysogenic variety as well as it could receive the other genophores and segregate the immune-2 variety.

If this explanation is correct we may anticipate the results of some cross-experiments. For instance we may expect that in all crosses in which the F_- parent is immune-2 or immune 1 and 2 the λ-genophore will not be received by this parent. In other words we will expect that the cross F_+ <u>lysogene</u> x F_- <u>immune 1 and 2</u> will <u>not</u> segregate a <u>lysogen-immune 2</u> variety and the cross F_+ sensitive x F_- immune 1 and 2 will not segregate an <u>immune-2</u> variety; neither will the Gal genes be carried from the F_+ to the F_- parent in any

of these cases.

Moreover, we may expect that F_+ immune 1 bacteria can probably produce, instead of a λ-phage, a non virulent λ^r genophore carring the gene $L_{pl}{}^r$, instead of $L_{pl}{}^+$. If this is true the cross F_+ immune 1 and 2 x F_- sensitive would segregate not only the variety immune 2 but also the variety immune 1.

Likewise we must take into consideration the possibility that a λ^S non virulent genophore may exist carring the gene $L_{pl}{}^S$. But this genophore could be uneasy to detect if not received by F_- immune 1. It is possible that the λ^r and λ^S genophores are unable to enter other bacteria by themselves and must be introduced during conjugation. This could be the reason for their non virulens as well as for the non virulens of many other genophores transformed into intermediaries in the crossing mechanism of bacteria and unable to act as parasites.

If this is the reason for the non virulens of the phages λ^r and λ^S, then we may expect that the characters $L_{pl}{}^r$ (immunity 1) and L_{pl} (sensitivity 1) can not be transduced by filtrate from F_+ to F_- bacteria. This could be an experimental way to test our assumptions concerning λ^r and λ^S.

We can not trust that F_- bacteria can be true lysogenes because we would expect that the production of λ-genophores, as well as the production of all other genophores, should be a property of F_+ bacteria. But if they can, then several other crossing experiments would be possible, the results of which could also be predicted on the bases of the genophore-hypotheses.

We may however, warn that some kind of fictitious lysogeny may occur in F_- bacteria carring the $L_{pl}{}^+$ gene if the bacteria by coningation are transformed into F_+.

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